

TRANSFORMATION IN E.coli DH5alpha

1. Thaw competent cells on ice. These can be prepared using the CaCl₂ protocol.
2. Add 1-2 μ l of DNA into the competent cells. Stir with a pipette tip a few times, then put right back on ice.
3. Incubate the cells on ice for 10 min.
4. Heat shock at 42 °C for exactly 90 seconds.
5. Return to ice for 90 seconds.
6. Add 800 μ L LB medium.
7. Incubate at 37 °C for 45-60 minutes in a water bath.
8. Plate the suspension on a LB agar plate containing the appropriate antibiotic.
9. Incubate plates upside down overnight at 37 °C.